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## INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

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<b>(21) International Application Number:</b> PCT/IN99/00011 <b>(22) International Filing Date:</b> 31 March 1999 (31.03.99) <b>(30) Priority Data:</b> 827/Mas/98 17 April 1998 (17.04.98) IN <b>(71)(72) Applicants and Inventors:</b> SUBBA RAO, Pillarisetti, Venkata [IN/IN]; 41/3, 13th Cross, Malleswaram, Bangalore 560 003 (IN). ANNADURAI, Ramasamy, Sambasivam [IN/IN]; 1840, 10th Main Road, 34th Cross, Banshankari II Stage, Bangalore 560 070 (IN). SRINIVAS, Malladi [IN/IN]; 319, IC Cross, II Phase, 6th Block, Banshankari III Stage, Bangalore 560 085 (IN). <b>(74) Agents:</b> ANAND, Pravin et al.; Anand & Anand Advocates, B-41, Nizammuddin East, New Delhi 110 013 (IN).		<b>(81) Designated States:</b> AE, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ, DE, DK, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, UA, UG, UZ, VN, YU, ZA, ZW, ARIPO patent (GH, GM, KE, LS, MW, SD, SL, SZ, UG, ZW), Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG).  <b>Published</b> <i>With international search report.</i>
<b>(54) Title:</b> AN ENVIRONMENT FRIENDLY ACARICIDE FORMULATION  <b>(57) Abstract</b>  This invention relates to an environment friendly acaricide formulation composition for the control of house dust mites comprising: plant derived acaricidal agent 0.01–0.1 % wt./vol.; plant derived disinfectant agent 0.3–3 % wt./vol.; plant derived protein denaturan 0.1–2 % wt./vol.; fungistat agent 0.1–3 % wt./vol.; dispersing agent (alcohol) 99.69–91.9 % wt./vol.		

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### *An Environment Friendly Acaricide Formulation*

This invention relates to an environment friendly acaricide formulation for the control of house dust mite population in the domestic environment responsible for high incidence of respiratory allergies and process of preparing the same.

#### 5 BACKGROUND

The major culprits for dust allergy are mites (*Dermatophagoides farinae* and *D. pteromyssinus*) prevalent in house dust. Universally, dust mites are minute co-inhabitants in almost every household and can not be seen with the naked eye. They are found in almost all home furnishing textiles and their favourite places are  
10 mattresses, cushions, carpets, upholstery and soft toys. The reactions in hypersensitive people range from itchy and watery eyes, repeated sneezes and running nose, cough and bronchial asthma to childhood eczema. The dust on which they thrive may comprise cotton, wool lint, animal and human dander, crumbs, pollens, molds, etc.

15 House dust mites principally feed on human scales which are primarily found in mattresses. Bedding, carpets etc. During occupation, the temperature and humidity of the human body provides an ideal microclimate in the mattresses for dust mites. Development from egg through larva, protonymph, tritonymph to adult requires about a month in cultures, under optimum conditions. An adult mite can  
20 live up to three months. Their food comprises of protein particles and fungi present in the dust.

Mites may occasionally become airborne during bed-making. It has also been demonstrated that they secrete or release some allergens during bed-making. The allergen may comprise mites, eggs, dead mites and their excreta. A gram of dust  
25 mite may contain up to 1000 mites.

Most particles of the faeces, whose physical properties are similar to pollen are deposited on the nasal mucosa and carried to the lungs causing localized inflammatory responses because of the high concentration of allergen.

Control of mite population in the domestic environment is the best method of preventing house dust allergy. The degree of cleanliness determines the number of house dust mites and the allergen level. Common control measures include vacuum cleaning, treating the carpets and bed spreads with insecticides, acaricides and fungicides. Reducing the mite population by interfering with the food chain has also been practised. However, a safe, environment friendly and effective formulation based on natural products for the control of house dust mite is not yet commercially available.

### PRIOR ART

A few formulations are commercially available like Acardust, Acarosan, Allerbiocid etc., containing benzyl benzoate, the chief acaricide agent in these formulation is toxic at higher concentrations to humans as well a pets. As the effective concentration of benzyl benzoate used in these formulations is very high, its wide spread use as a domestic acaricide could be harmful.

In one acaricidal formulation, derivatives of phenols in combination with several natural oils have been used as the active acaricide agent in combination with an antibiotic Natamycin as a fungicide. But the wide spread use of phenolic derivatives and essential oils is not safe from physiological and odour point of view. Moreover, the fungicide as such can not destroy the mites.

Apart from this, a few chemicals like benzyl alcohol, primiphos methyl, dibutyl phthalate, gama-hexachlorocyclohexane and diethyl-m-toluamide have been reported in literature as miticides. But from the toxicology and environmental safety point of view their use is not recommended.

Accordingly, the **object** of this invention is to provide an environment friendly acaricide formulation for domestic use which should have the following characteristics:

- All the chemicals used should be safe from the toxicology point of view.

- Should have multiple modes of action i.e., it should control the mite population, prevent the growth of fungi, reduce the existing allergen levels, act as a disinfectant as well as prevent the mites from developing resistance to these chemicals.

5                   - Should not have an offensive odour.

Further, re-establishment of house dust mites after treatment with acaricides is the common problem due to the existence of nymph and eggs. Moreover, the miticide cannot reach the deeper layers of carpets and upholstery. Accordingly, the second **object** of this invention is to control the mites and prevent its re-  
10 establishment by preparing the composition which can not only kill the adult mites but also be a ovicide and a larvicide.

To achieve the said objectives this invention provides an environment friendly acaricide formulation for the control of house dust mites comprising :

- |    |                                    |                       |
|----|------------------------------------|-----------------------|
| 15 | - plant derived acaricidal agent   | - 0.01-0.1 % wt./vol. |
|    | - plant derived disinfectant agent | - 0.1-3 % wt./vol.    |
|    | - plant derived protein denaturant | - 0.1-2 % wt./vol.    |
|    | - fungistat agent                  | - 0.1-3% wt./vol.     |
|    | - dispersing agent (alcohol)       | - 99.69-91.9% wt./vol |

The plant derived acaricidal agent is neem seed kernel extract containing  
20 azadirachtin / azadirachtin A of 2-90 % enrichment and preferably of 20-35 % enrichment.

The neem seed kernel extract contains limonoids like nimbin, salannin, desacetylnimbin, desacetylsalannin, nimbandiol, azadirachtin-B and salannolacetate for preventing the mites from developing resistance against the  
25 active ingredient.

The plant derived disinfectant agent is an alcoholic extract of resins like stryax benzoin and the plant derived protein denaturant is plant polyphenols like

tannic acid, condensed tannins, phenolic compounds like gallic acid and phloroglucinol.

The fungistat agents are fungicides used in food industry like natamycin, nipagin and the dispersing agents are ethanol, methanol and isopropyl alcohol.

5 The ingredients viz. plant derived acaricidal agent, plant derived disinfectant agent, plant derived protein denaturant and fungistat agent of this composition are solids which are dissolved in an alcoholic solvent (dispersing agent) to give a clear pale brown coloured solution.

The invention will now be described with reference to the following  
10 examples.

#### EXAMPLE – 1

S. No.	Ingredients	Weight/volume (%)
1.	Neem seed kernel extract containing azadirachtin of 20% enrichment	0.1
2.	Alcoholic extract of benzoin resin	3.0
3.	Tannic acid	1.0
4.	Nipagin	1.0
5.	Ethanol	94.9

#### EXAMPLE – 2

15

S. No.	Ingredients	Weight/volume (%)
1.	Neem seed kernel extract containing azadirachtin of 35% enrichment	0.1
2.	Alcoholic extract of benzoin resin	3.0
3.	Tannic acid	1.0
4.	Nipagin	1.0
5.	Ethanol	94.9

**EXAMPLE – 3**

S. No.	Ingredients	Weight/volume (%)
1.	Neem seed kernel extract containing azadirachtin of 90% enrichment	0.1
2.	Alcoholic extract of benzoin resin	3.0
3.	Tannic acid	1.0
4.	Nipagin	1.0
5.	Ethanol	94.9

**EXAMPLE – 4**

5

S. No.	Ingredients	Weight/volume (%)
1.	Neem seed kernel extract containing azadirachtin of 2% enrichment	0.1
2.	Alcoholic extract of benzoin resin	3.0
3.	Tannic acid	1.0
4.	Nipagin	1.0
5.	Ethanol	94.9

**EXAMPLE – 5**

S. No.	Ingredients	Weight/volume (%)
1.	Neem seed kernel extract containing azadirachtin of 20% enrichment	0.1
2.	Alcoholic extract of benzoin resin	3.0
3.	Tannic acid	1.0
4.	Nipagin	1.0
5.	Isopropyl alcohol	94.9

**EXAMPLE – 6**

S. No.	Ingredients	Weight/volume (%)
1.	Neem seed kernel extract containing azadirachtin of 35% enrichment	0.1
2.	Alcoholic extract of benzoin resin	3.0
3.	Tannic acid	1.0
4.	Nipagin	1.0
5.	Isopropyl alcohol	94.9

**EXAMPLE – 7**

5

S. No.	Ingredients	Weight/volume (%)
1.	Neem seed kernel extract containing azadirachtin of 90% enrichment	0.1
2.	Alcoholic extract of benzoin resin	3.0
3.	Tannic acid	1.0
4.	Nipagin	1.0
5.	Isopropyl alcohol	94.9

**EXAMPLE – 8**

S. No.	Ingredients	Weight/volume (%)
1.	Neem seed kernel extract containing azadirachtin of 2% enrichment	0.1
2.	Alcoholic extract of benzoin resin	3.0
3.	Tannic acid	1.0
4.	Nipagin	1.0
5.	Isopropyl alcohol	94.9



**Conclusion:**

Composition in Table 2 is very efficient in terms of speed of action and mite elimination. The total adult population is immobilized within an hour of treatment. A biweekly spray of 200  $\mu$ l/100mg of the culture is required for 8 weeks to  
5 completely eliminate the population. Reestablishment on treated areas is totally prevented after 8 weeks. After eradication a biweekly prophylactic spray can contain population build up.

**We claim:**

1. An environment friendly acaricide formulation composition characterized by:
  - plant derived acaricidal agent - 0.01-0.1 % wt./vol.
  - plant derived disinfectant agent - 0.1-3 % wt./vol.
  - 5 - plant derived protein denaturant - 0.1-2 % wt./vol.
  - fungistat agent - 0.1-3% wt./vol.
  - dispersing agent (alcohol) - 99.69-91.9% wt./vol
2. Formulation as claimed in claim 1 wherein the plant derived acaricidal agent is neem seed kernel extract containing azadirachtin of 2-90 % enrichment.
- 10 3. Formulation as claimed in claim 1 wherein the plant derived acaricidal agent is neem seed kernel extract containing azadirachtin A of 2-90 % enrichment.
4. Formulation composition as claimed in claim 1 wherein the plant derived acaricidal agent is neem seed kernel extract containing azadirachtin / azadirachtin A preferably of 20-35 % enrichment.
- 15 5. Formulation as claimed in claim 2 wherein the neem seed kernel extract contains limonoids like nimbin, salannin, desacetylnimbin, desacetylsalannin, nimbandiol, azadirachtin-B and salannolacetate for preventing the mites from developing resistance against the active ingredient.
6. Formulation as claimed in claim 1 wherein the plant derived disinfectant  
20 agent is an alcoholic extract of resins like stryax benzoin.
7. Formulation as claimed in claim 1 wherein the plant derived protein denaturant is plant polyphenols like tannic acid, condensed tannins, phenolic compounds like gallic acid and phloroglucinol.
8. Formulation as claimed in claim 1 wherein the fungistat agents are  
25 fungicides used in food industry like natamycin, nipagin.
9. Formulation as claimed in claim 1 wherein the dispersing agents are ethanol, methanol, isopropyl alcohol.

# INTERNATIONAL SEARCH REPORT

International application No.  
PCT/IN 99/00011

## A. CLASSIFICATION OF SUBJECT MATTER

IPC<sup>6</sup>: A 01 N 65/00

According to International Patent Classification (IPC) or to both national classification and IPC

## B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

IPC<sup>6</sup>: A 01 N

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## C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	US 5 405 612 A (LOCKE et al.), 11 April 1995 (11.04.95), claims.	1-5
A	EP 0 405 291 A1 (W.R.GRACE & CO.), 02 January 1991 (02.01.91), example 5; claims 8,9.	1-5,9
A	DATABASE WPI ON EPOQUE, week 9408, London: Derwent Publications Ltd., AN 94-061960, class B 04m JP 6016515 A (NIPPON KAYAKU KK), abstract.	1-5
A	DE 195 32 447 A1 (REMBOLD), 06 March 1997 (06.03.97), example 1; claims 1-5,15,17,18.	1-5
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☐ Further documents are listed in the continuation of Box C.

☒ See patent family annex.

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US A	5405612	11-04-1995		AT E	111307	15-09-1994	
				AU A1	62627/90	04-07-1991	
				AU B2	633622	04-02-1993	
				CA AA	2013754	26-06-1991	
				DE C0	69012538	20-10-1994	
				DE T2	69012538	12-01-1995	
				EP B1	436257	10-07-1991	
				EP B1	436257	14-09-1994	
				ES T3	2060004	16-11-1994	
				JP A2	4364103	16-12-1992	
				NZ A	236580	28-04-1992	
				US A	5356628	18-10-1994	
				US A	5368856	29-11-1994	
				US A	5411736	02-05-1995	
EP A1	405291	02-01-1991		AU A1	57759/90	03-01-1991	
				AU B2	619720	30-01-1992	
				CA AA	2016964	26-12-1990	
				DE C0	69011988	06-10-1994	
				DE T2	69011988	12-01-1995	
				DK T3	405291	03-10-1994	
				EP B1	405291	31-08-1994	
				ES T3	2058686	01-11-1994	
				JP A2	3038506	19-02-1991	
				JP B2	2841746	24-12-1998	
				NZ A	234226	25-06-1992	
				PT A	94495	08-02-1991	
				PT B	94495	28-02-1997	
				US A	5001146	19-03-1991	
				US A	5124349	23-06-1992	
				US B1	5124349	20-10-1998	
JP A2	6016515	25-01-1994		keine - none - rien			
DE A1	19532447	06-03-1997		keine - none - rien			